



MONOCLONAL ANTIBODY

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Catalog No. KMU-M01

Anti-Nitroguanosine monoclonal antibody

BACKGROUND

8-Nitroguanosine is a nitrated nucleic acid which is formed by peroxyxynitrite, myeloperoxidase, nitrite, and peroxide. It is known that the nitration of guanine is enhanced in virus infection^{1, 2}, bacterial infection^{3, 4}, inflammatory disease⁵, cancers⁵, and diseases associated with smoking⁶. 8-nitroguanosine is thought to be one of the makers of DNA damage caused by oxidative stress. Cyclic GMP (cGMP) is one of the important substances for the signal transfer. On the other hand, 8-Nitro-cGMP (nitrated cGMP) has been identified *in vivo*³. Therefore, 8-Nitro-cGMP can potentially act as a mediator for reactive oxygen signaling^{3, 7-9}. Although the product, Anti-Nitroguanosine monoclonal antibody (NO₂G52), does not cross-react with normal nucleotide bases, it selectively reacts with nitro functionalized nucleotides such as nitro-cGMP, nitro-GMP, and nitro-GTP. Therefore, Anti-Nitroguanosine polyclonal antibody is universal antibody of nitrated guanine which modified 8th position of guanine with nitro group. NO₂G52 is commonly used for immunostaining. In addition, NO₂G52 can potentially utilized for affinity purification of nitroguanine derivative^{3, 6}.

Product type	Primary antibodies
Host	Mouse (BALB/c)
Form	Liquid 1 mg/ml PBS solution; 0.1% ProClin as a preservative
Volume	200 ug
Clone	NO ₂ G52
Isotype	IgG1

Application notes	IHC, ELISA Immunohistochemistry, 10 ug/ml ELISA, 1 ug/ml Optimal dilutions/concentrations should be determined by the end user.
Cross reactivity	Strongly reacts (10 umol/l) 8-NO ₂ -guanosine 8-NO ₂ -guanine Slightly cross-reacts (>1 mmol/l) 8-Br-guanosine 8-Br-guanine 8-Cl-guanine



No cross-reaction

guanosine, guanine, 8-OH-guanine, 8-OH-deoxyguanosine, xanthine, adenine, adenosine, thymine, deoxythymidine, uracil, uridine, 3-NO₂-tyrosine, 2-NO₂-imidazole, cytosine

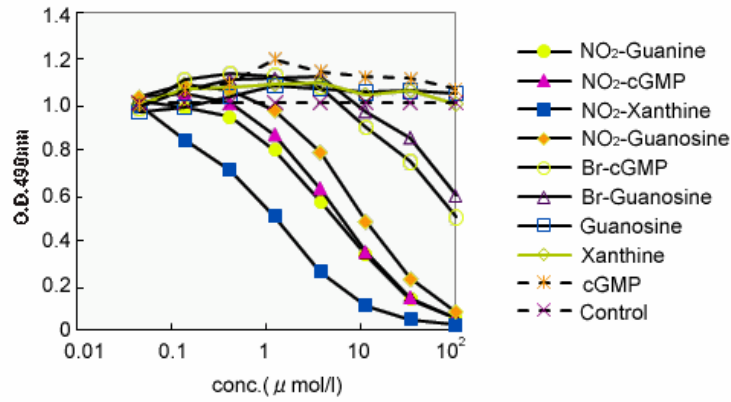


Fig. 1. The reactivity of Monoclonal Antibody(NO₂G52).

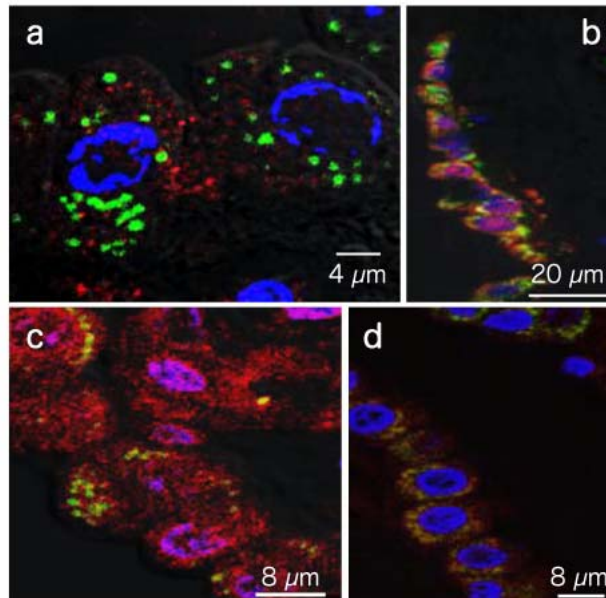


Fig. 2. Production of 8-nitroguanine by airway epithelial cell with Idiopathic Fibrosis (IPS).

A. 8-nitroguanine (green) and iNOS (red) **B.** Bright-field **C.** 8-nitroguanine (green) and 8-oxoguanine (Red) **D.** 8-nitroguanine (green) and mitochondria (red)

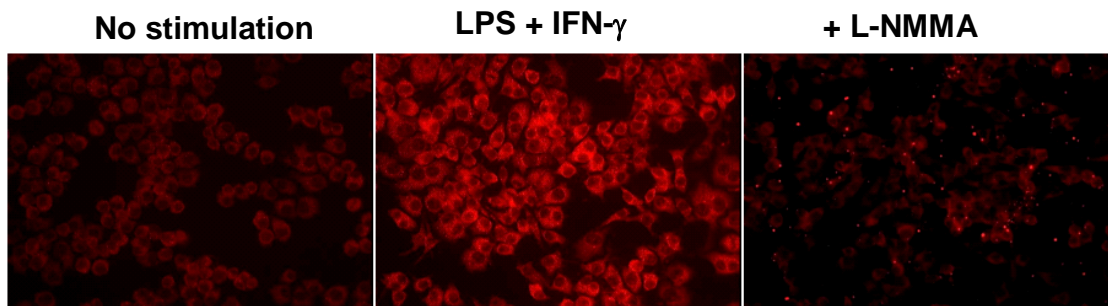


Fig. 3. Endogenous guanine nitration in RAW 264.7 cells, a murine macrophage cell line



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Storage

Store below -20°C (below -70°C for prolonged storage).

Aliquot to avoid cycles of freeze/thaw.

References

- 1) T. Akaike, S. Okamoto, T. Sawa, J. Yoshitake, F. Tamura, K. Ichimori, K. Miyazaki, K. Sasamoto and H. Maeda, 8-nitroguanosine formation in viral pneumonia and its implication for pathogenesis, *Proc. Natl. Acad. Sci. USA*, **100**, 685-690 (2003).
- 2) J. Yoshitake, T. Akaike, T. Akuta, F. Tamura, T. Ogura, H. Esumi, and H. Maeda, Nitric oxide as an endogenous mutagen for Sendai virus without antiviral activity, *J. Virol.*, **78**, 8709-8719 (2004).
- 3) T. Sawa, M. H. Zaki, T. Okamoto, T. Akuta, Y. Tokutomi, S. Kim-Mitsuyama, H. Ihara, A. Kobayashi, M. Yamamoto, S. Fujii, H. Arimoto, and T. Akaike, Protein S-guanylation by the biological signal 8-nitroguanosine 3',5'-cyclic monophosphate, *Nat. Chem. Biol.*, **3**, 727-735 (2007).
- 4) M. H.Zaki, S. Fujii, T. Okamoto, S. Islam, S. Khan, K. A. Ahmed, T. Sawa, and T. Akaike, Cytoprotective function of heme oxygenase 1 induced by a nitrated cyclic nucleotide formed during murine salmonellosis, *J. Immunol.*, **182**, 3746-3756 (2009).
- 5) Y. Terasaki, T. Akuta, M. Terasaki, T. Sawa, T. Mori, T. Okamoto, M. Ozaki, M. Takeya and T. Akaike, Guanine nitration in idiopathic pulmonary fibrosis and its implication for carcinogenesis, *Am. J. Respir. Crit. Care. Med.*, **174**, 665-673 (2006).
- 6) T. Sawa, M. Tatemichi, T. Akaike, A. Barbin and H. Ohshima, Analysis of urinary 8-nitroguanine, a marker of nitrative nucleic acid damage, by high-performance liquid chromatography-electrochemical detection coupled with immunoaffinity purification: association with cigarette smoking, *Free Radic. Biol. Med.*, **40**, 711-720 (2006).
- 7) M. Feelisch, Nitrated cyclic GMP as a new cellular signal, *Nat. Chem. Biol.*, **3**, 687-688 (2007).
- 8) K. A. Ahmed, T. Sawa, T. Akaike, Protein cysteine S-guanylation and electrophilic signal transduction by endogeneous nitro-nucleotides, *Amino Acids*, *in press* (2010).
- 9) T. Sawa, H. Arimoto and T. Akaike, Regulation of redox signaling involving chemical conjugation of protein thiols by nitric oxide and electrophiles, *Bioconjugate Chem.*, *in press* (2010).

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TOYO 2CHOME, KOTO-KU, TOKYO, 135-0016, JAPAN

URL: <http://www.cosmobio.co.jp>

e-mail: export@cosmobio.co.jp

[Outside Japan] Phone : +81-3-5632-9617

[国内連絡先] Phone : +81-3-5632-9610

FAX : +81-3-5632-9618

FAX : +81-3-5632-9619